

THAT WHICH IS CLAIMED IS:

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1. A method of hydrolyzing a galactose-containing oligosaccharide present in a substrate, comprising:
contacting the substrate with a hyperthermophilic α -galactosidase isolated from the group consisting of *Thermotoga maritima*, *Thermotoga elfii*, and *Thermotoga* sp. T2; and
heating the substrate to a temperature at which the hyperthermophilic α -galactosidase is active, for a period of time sufficient to hydrolyze the oligosaccharide.
2. The method of Claim 1, wherein the oligosaccharide is selected from the group consisting of raffinose, stachyose and verbascose.
3. The method of Claim 1, wherein the substrate is animal feed.
4. The method of Claim 1, wherein the substrate is soybean meal.
5. The method of Claim 1, wherein the substrate is human food.
6. The method of Claim 1, wherein the hyperthermophilic α -galactosidase is isolated from *Thermotoga maritima*.
7. The method of Claim 1, wherein the hyperthermophilic α -galactosidase is isolated from *Thermotoga maritima* DSM3109.
8. The method of Claim 1, wherein the oligosaccharide is hydrolyzed into galactose monomers.
9. The method of Claim 1, wherein the method is carried out under conditions of 70% moisture.

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See 15 page
for details
of the method

10. The method of Claim 1, wherein the method is carried out under conditions of 25% moisture.

11. The method of Claim 1, wherein the heating occurs at 80°C.

12. The method of Claim 1, wherein the heating occurs at 85°C.

13. The method of Claim 1, wherein the heating occurs at 90°C.

14. The method of Claim 1, wherein the heating occurs at 100°C.

15. The method of Claim 1, wherein the hyperthermophilic α -galactosidase is produced by:

(a) culturing a host cell comprising an expression vector containing a polynucleotide sequence encoding an hyperthermophilic α -galactosidase;

(b) expressing the hyperthermophilic α -galactosidase; and

(c) recovering the hyperthermophilic α -galactosidase from the host cell culture.

16. The method of Claim 15, wherein the polynucleotide has the sequence of **SEQ ID NO:1**.

17. The method of Claim 15, wherein the polynucleotide is selected from the group consisting of

(a) DNA having the nucleotide sequence of **SEQ ID NO:1**;

(b) polynucleotides that encode an hyperthermophilic α -galactosidase and hybridize to DNA of (a) above under stringent conditions; and

(c) polynucleotides that encode an hyperthermophilic α -galactosidase and differ from the DNA of (a) or (b) above due to the degeneracy of the genetic code.

18. The method according to Claim 15 wherein the polynucleotide encodes an hyperthermophilic α -galactosidase having the amino acid sequence of **SEQ ID NO:2**.

19. A method of preparing an animal feed composition comprising a hydrolyzed galactose-containing oligosaccharide, comprising:

contacting ingredients of the animal feed composition with a hyperthermophilic α -galactosidase during the processing of the animal feed, wherein the hyperthermophilic α -galactosidase is contacted with the animal feed ingredients prior to a heating step in the animal feed processing for a period of time sufficient to allow the hyperthermophilic α -galactosidase to hydrolyze the galactose-containing oligosaccharide; and

wherein the hyperthermophilic α -galactosidase is isolated from the group consisting of *Thermotoga maritima*, *Thermotoga elfii*, and *Thermotoga* sp. T2.

20. The method of Claim 19, wherein said galactose-containing oligosaccharide is selected from the group consisting of raffinose, stachyose and verbascose.

21. The method of Claim 19, wherein the animal feed comprises soybean meal.

22. The method of Claim 19, wherein the animal feed comprises soybean flakes.

23. The method of Claim 19, wherein the animal feed is chicken feed.

24. The method of Claim 19, wherein the hyperthermophilic α -galactosidase is isolated from *Thermotoga maritima*.

25. The method of Claim 19, wherein the hyperthermophilic α -galactosidase is isolated from *Thermotoga maritima* DSM3109.

26. The method of Claim 19, wherein the oligosaccharide is hydrolyzed into galactose monomers.

27. The method of Claim 19, wherein the contacting of the hyperthermophilic α -galactosidase with the ingredients of the animal feed composition is carried out under conditions of 70% moisture.

28. The method of Claim 19, wherein the contacting of the hyperthermophilic α -galactosidase with the ingredients of the animal feed composition is carried out under conditions of 25% moisture.

29. The method of Claim 19, wherein the contacting of the hyperthermophilic α -galactosidase with the ingredients of the animal feed composition is carried out under conditions of 45% moisture.

30. The method of Claim 19, wherein the heating step occurs at 80°C.

31. The method of Claim 19, wherein the heating step occurs at 85°C.

32. The method of Claim 19, wherein the heating step occurs at 90°C.

33. The method of Claim 19, wherein the heating step occurs at 100°C.

34. The method of Claim 19, wherein the contacting of the ingredients of the animal feed composition with the hyperthermophilic α -

galactosidase occurs prior to a final pelleting step in the animal feed processing.

35. The method of Claim 19, wherein the hyperthermophilic α -galactosidase is produced by:

- (a) culturing a host cell comprising an expression vector containing a polynucleotide sequence encoding an hyperthermophilic α -galactosidase;
- (b) expressing the hyperthermophilic α -galactosidase; and
- (c) recovering the hyperthermophilic α -galactosidase from the host cell culture.

36. The method of Claim 35, wherein the polynucleotide has the sequence of **SEQ ID NO:1**.

37. The method of Claim 35, wherein the polynucleotide is selected from the group consisting of

- (a) DNA having the nucleotide sequence of **SEQ ID NO:1**;
- (b) polynucleotides that encode an hyperthermophilic α -galactosidase and hybridize to DNA of (a) above under stringent conditions; and
- (c) polynucleotides that encode an hyperthermophilic α -galactosidase and differ from the DNA of (a) or (b) above due to the degeneracy of the genetic code.

38. The method according to Claim 35 wherein the polynucleotide encodes an hyperthermophilic α -galactosidase having the amino acid sequence of **SEQ ID NO:2**.

39. The method according to Claim 19, wherein the hyperthermophilic α -galactosidase is in liquid solution when the hyperthermophilic α -galactosidase is contacted with the ingredients of the animal feed composition.

40. The method according to Claim 19, wherein the hyperthermophilic α -galactosidase is in dried form when the hyperthermophilic α -galactosidase is contacted with the ingredients of the animal feed composition.

41. The method according to Claim 19, wherein the hyperthermophilic α -galactosidase is partially purified when the hyperthermophilic α -galactosidase is contacted with the ingredients of the animal feed composition.

42. The method according to Claim 19, wherein the hyperthermophilic α -galactosidase is in substantially purified form when the hyperthermophilic α -galactosidase is contacted with the ingredients of the animal feed composition.

43. An animal feed produced according to the method of Claim 19.

44. A food additive for the reduction of gastrointestinal distress in mammals, comprising a hyperthermophilic α -galactosidase isolated from the group consisting of *Thermotoga maritima*, *Thermotoga elfii*, and *Thermotoga* sp. T2.

same as
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+
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excipient?

45. The food additive of Claim 44, wherein the hyperthermophilic α -galactosidase is isolated from *Thermotoga maritima*.

46. The food additive of Claim 44, wherein the hyperthermophilic α -galactosidase is isolated from *Thermotoga maritima* DSM3109.

47. The food additive of Claim 44, wherein the hyperthermophilic α -galactosidase is produced by:

(a) culturing a host cell comprising an expression vector containing a polynucleotide sequence encoding an hyperthermophilic α -galactosidase;

(b) expressing the hyperthermophilic α -galactosidase; and

(c) recovering the hyperthermophilic α -galactosidase from the host cell culture.

48. The food additive of Claim 47, wherein the polynucleotide has the sequence of **SEQ ID NO:1**.

49. The food additive of Claim 47, wherein the polynucleotide is selected from the group consisting of

- (a) DNA having the nucleotide sequence of **SEQ ID NO:1**;
- (b) polynucleotides that encode an hyperthermophilic α -galactosidase and hybridize to DNA of (a) above under stringent conditions; and
- (c) polynucleotides that encode an hyperthermophilic α -galactosidase and differ from the DNA of (a) or (b) above due to the degeneracy of the genetic code.

50. The food additive according to Claim 47 wherein the polynucleotide encodes an hyperthermophilic α -galactosidase having the amino acid sequence of **SEQ ID NO:2**.

51. A method of preventing gastrointestinal distress in a mammal, wherein the gastrointestinal distress is caused by food containing at least one oligosaccharide selected from the group consisting of raffinose, stachyose and verbascose, comprising:

contacting the food with a hyperthermophilic α -galactosidase isolated from the group consisting of *Thermotoga maritima*, *Thermotoga elfii*, and *Thermotoga* sp. T2; and then
heating the food for a period of time sufficient to allow the hyperthermophilic α -galactosidase to hydrolyze the oligosaccharide.

52. A processing additive for the removal of galactose-containing oligosaccharides in a process of making edible soybean protein, comprising a hyperthermophilic α -galactosidase isolated from the group consisting of *Thermotoga maritima*, *Thermotoga elfii*, and *Thermotoga* sp. T2.

same as claim 44

53. A method of removing galactose-containing oligosaccharides from a soybean substrate being processed to produce an edible soybean protein, comprising:

contacting the soybean substrate with a hyperthermophilic α -galactosidase isolated from the group consisting of *Thermotoga maritima*, *Thermotoga elfii*, and *Thermotoga* sp. T2;

heating the soybean substrate at a temperature and for a length of time sufficient to hydrolyze the galactose-containing oligosaccharides; and

removing the hydrolyzed galactose-containing oligosaccharides from the soybean substrate prior to a final extraction or fractionation of the edible soybean protein.

54. The method of Claim 53, wherein the heating occurs prior to the removal of oil from the soybean substrate.

55. The method of Claim 53, wherein the heating occurs after the removal of oil from the soybean substrate.

56. The method of Claim 53, wherein the soybean substrate is soybean flakes.

57. An isolated edible soybean protein produced by the method of Claim 53.

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A4